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Biocatalytic processes have become very important for the chemical industry. In this connection, carrying out chemical reactions with the aid of biological catalysts is particularly interesting in those fields of application in which it is possible to utilize the frequently found enzyme property of preferably converting or forming in chemical reactions with chiral or prochiral components one of the two enantiomers.

between lines 19 and 20, insert the following new heading:

A3

2. Description of the Related Art

Page 2, between lines 30 and 31, insert the following new heading:

AH

Summary of the Invention

Page 4, between lines 14 and 15, insert the following:

Brief Description of the Drawings



Figure 1 shows the nucleotide sequence (SEQ ID No. 19) of the *Prunus* amygdalus HNL5 gene obtained by PCR amplification. The start codon (ATG) and stop codon of the open reading frame are printed in bold type, and the nucleotides in the intron regions are indicated in lower case letters. The peripheral sequences which have been attached via the PCR primers and which are not part of the *HNL5* gene are underlined. The splice sites of the introns were identified with the aid of the consensus sequence "GT. . . . AG".

Figure 2 shows an intron-free *Prunus amygdalus HNL5* gene being obtained by PCR splicing. By means of a specific PCR strategy using overlapping primers, the coding regions were linked to one another by 4 successive PCR reactions.

/ **>**  Figure 3 shows the amino acid sequence (SEQ ID No. 20) of the *Prunus* amygdalus hydroxynitrile lyase (HNL5), derived from the nucleotide sequence of the *HNL5* gene (SEQ ID No. 19). The signal sequence determined from sequence analysis is printed in bold type and the postulated processing site is indicated by an arrow. Possible glycosylation sites (PROSITE patterns) are underlined.

Figure 4 shows the nucleic acid sequence (SEQ ID No. 21) of the DNA fragment coding for a secretory hybrid protein (PamHNL5xGOX) with HNL activity, consisting of sequences of the *Prunus amygdalus HNL5* gene and the *Aspergillus niger* glucose oxidase gene.

Figure 5 shows the amino acid sequence (SEQ ID No. 22) of the hybrid protein PamHNL5xGOX, derived from the nucleic acid sequence (SEQ ID No. 21) of Figure 4.

Figure 6 shows the comparison of the amino acid sequences of *Prunus amygdalus* HNL5 (SEQ ID No. 20) and of the hybrid protein PamHNL5xGOX (SEQ ID No. 22). Sequence parts of *Aspergillus niger* glucose oxidase are underlined. Sequence regions having no significant homology between the two proteins are printed in italics, and the signal peptides are printed in bold type.

Figure 7 shows the analysis of HNL preparations by SDS PAGE. Details are described in Example 11.

Figure 8 shows the nucleotide sequence (SEQ ID No. 23) of the *Prunus* amygdalus HNL1 gene obtained by PCR amplification.



Figure 9 shows the amino acid sequence (SEQ ID No. 24) of *Prunus amygdalus* hydroxynitrile lyase (HNL1), derived from the nucleotide sequence of the *HNL1* gene (SEQ ID No. 23).

### Description of the Preferred Embodiments

Page 10, below the last line of the page, insert the following paragraph:



The present invention will now be described in more detail by the following Examples. However, the following Examples are merely illustrative in nature. Other Examples within the scope of the claims are also possible. Thus, the following should not be construed to limit the spirit and scope of the claims.

# Page 12, replace the paragraph beginning at line 1 with the following paragraph:



Since it was known that a plurality of hydroxynitrile lyase isoenzymes whose sequences are highly homologous to one another can appear in *Prumus species* (Hu and Poulton, 1999), gene-specific PCR primers based on sequence homology of the *Prunus serontina mdl5* gene and the *Prunus amygdalus MDL1* gene (Suelves et al., 1998) were prepared:

Primer 1: 5'-CGGAATTCACAATATGGAGAAATCAACAATGTCAG-3' (SEQ ID No. 1)

Primer 2: 5'-CGGAATTCTTCACATGGACTCTTGAATATTATG-3' (SEQ ID No. 2).

Please replace page 16 with the attached substitute page 16.

# Page 17, replace the paragraph beginning at line 25 with the following paragraph:



Oligonucleotide primers:

PCRHNL5-a

5'-

TCGAATTCGAGCTCGGTACCCGGGGATCCTCTAGAAATAATTTTGTTTAACTTTA AGAAGGAGATATACATATGGAGAAATCAACAATGTCAGTTATACTATTTGTGTTG CATC -3' (SEQ ID No. 10)

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PCRHNL5-e

5'-

CGAATTCGCCCTTTCGCATGCTCACATGGACTCTTGAATATTATGAATAGCCTC-3'

(SEQ ID No. 11)

Page 23, replace the section between lines 20 and 25 with the following section:

Oligonucleotide primers used

**GLUCOX1** 

5'-

CACGAATTCATCATGCAGACTCTCCTTGTGAGCTCGCTTGTGGTCTCCCT

CGCTGCGGCCCTGCCACACTAC-3' (SEQ ID No. 12)



**GLUCOX2** 

5'-

TGCGGCCCTGCCACACTACATCAGGAGCAATGGCATTGAAGCCTACAACG

CCACTGATACAAGCTCGGAAGGATC-3' (SEQ ID No. 13)

**GLUCOXCT** 

5'-GAATTCGCATGCGCCGCTCACTGCATTGACCTTTCTTGCAGGATTTGAAG-3'

(SEQ ID No. 14)

## Page 25, replace the paragraph beginning at line 35 with the following paragraph:

A genomic DNA fragment having the coding region of the *Prunus amygdalus*HNL1 gene was amplified from genomic almond DNA (preparation, see example 1) with the aid of a PCR using primers mandlp2f (5'-ACTACGAATTCGACCATGGAGAAATCAAC-3') (SEQ ID No. 15) and ecpamHNL1e (5'-CAGAATTCGCCCTTGTGCATGCATCGATTAAAGAACCAAG GATGCTGCTGAC-3') (SEQ ID No. 16).

### Page 26, replace the paragraph beginning at line 28 with the following paragraph:

The DNA sequence of the *Prumus amygdalus HNL1* gene was verified and finally determined by amplifying another genomic DNA fragment using primers mand1p3f (5'-ACTACGAATTCGACCATGGAGAAATCAACAATG-3') (SEQ ID No. 17) and pamHNL1end (5'-ATGCTGCTGACTTGAGGGAATC-3') (SEQ ID No. 18). The amplification was carried out in 50 µl reactions with 2.5 units of "Hotstar" DNA polymerase (Qiagen GmbH, Hilden, Germany), in each case 10 pmol of the two primers, 2 µl of a dNTP mix (5 mM each) and 50 ng of genomic almond DNA in standard PCR buffer (Qiagen GmbH, Hilden, Germany). The following PCR program was used:

15 min 95°C, then 5 cycles of 1 min at 94°C, 30 sec at 55°C and 2 min at 72°C, then 30 cycles with 1 min at 94°C, 30 sec at 68°C and 2 min at 72°C and a final extension step at 72°C for 7 min.

#### In the Sequence Listing: